

A new species of South Texas scrubsnail, *Praticolella* (von Martens, 1892) (Gastropoda: Polygyridae)

Kathryn E. Perez¹

Eli Ruiz

Marco Martinez Cruz

Department of Biology
University of Texas Rio Grande Valley
1201 West University Drive
Edinburg, TX 78539 USA

Russell L. Minton

School of Science and Computer Engineering
University of Houston Clear Lake
2700 Bay Area Boulevard MC 39
Houston, TX 77058 USA

ABSTRACT

The *Praticolella* of South Texas are highly visible and abundant snails with a confusing taxonomic history. In this paper, we provide 16S mitochondrial rDNA and morphological evidence to distinguish a new species of *Praticolella*, *Praticolella salina*, from southernmost coastal Texas. This native species previously was considered a distinct race of *P. griseola*, which we demonstrate does not occur natively in Texas.

Additional Keywords: mtDNA, *Praticolella griseola*, *Praticolella mexicana*, Cameron County, Texas

INTRODUCTION

Praticolella (von Martens, 1892) are small (7–15 mm wide), globose, helicoid land snails found in open, grassy habitats. Two species in this genus, *P. griseola* (Pfeiffer, 1841) and *P. mexicana* Perez, 2011, have established populations worldwide via human-mediated transport (Robinson 1999, Perez 2011). *Praticolella* sensu stricto contains ten currently recognized species that occur in Texas. Six species are found in the Rio Grande Valley of South Texas, including four native and two non-native species of *Praticolella*. The South Texas *Praticolella* have a great deal of overlap in habitat, shell shape, color, aperture shape, and shell banding patterns; indeed, this region has been called a “great melting pot” for these snails (Cheatum and Fullington, 1971).

Over the last 150+ years, previous workers have recognized a unique population of *Praticolella* located in coastal South Texas, referring to it as a unique “race” of *P. griseola* (e.g. Orcutt, 1915; Rehder, 1966). In 2011, Perez described *Praticolella mexicana* and distinguished this species occurring in Texas from *P. griseola* and *P. berlandieriana* (Moricand, 1833). Phylogenetic work

by Perez (2011) based on mtDNA sequences established that a few individuals identified as *P. griseola* from Cameron County Texas formed a distinct clade; with only a single population represented in that study, that author declined to establish a formal distinctive taxonomic status for that population. This population was also found to be distinct using geometric morphometrics (Perez, 2011). In the present study, we sampled additional populations of *Praticolella* from coastal Cameron County, Texas, and used anatomical and genetic data to determine that these populations represented a previously unrecognized, distinct species.

MATERIALS AND METHODS

Collections and Molecular Methods: Representatives of the populations in Cameron County were collected by hand and individuals were frozen at -20°C prior to DNA extraction. We amplified the mitochondrial 16S rDNA gene of twenty individuals from four of these populations (Figure 1, Table 1) using the degenerate 16sar-deg and 16sbr-deg primers described in Perez (2011). Methods for DNA extraction and PCR also follow Perez (2011), and Sanger sequencing was carried out by Beckman Coulter Genomics. Contigs were assembled in SeqMan Pro (DNASTAR 2014, SeqMan Pro®. Madison, WI) and added to the sequences used in Perez (2011). The dataset was aligned using MUSCLE 3.7 (Edgar, 2004) followed by elimination of poorly aligned positions in Gblocks 0.91b (Castresana 2000) implemented at Phylogeny.fr (Dereeper et al. 2008) (<http://phylogeny.lirmm.fr/phylo.cgi/index.cgi>). We used jModeltest (2.1.7) (Guindon and Gascuel, 2003; Darriba, et al. 2012) to select TIM1+I+G (Posada 2003) as the best model for our data. Maximum likelihood analysis and 1000 bootstrap replicates were carried out in Garli 2.01 (Zwickl, 2006). Base frequencies and substitution rate categories were estimated from the data.

Species Delimitation Analyses: We used three methods to assess whether our labeled clades represented

¹ Author for correspondence: perezke@gmail.com.

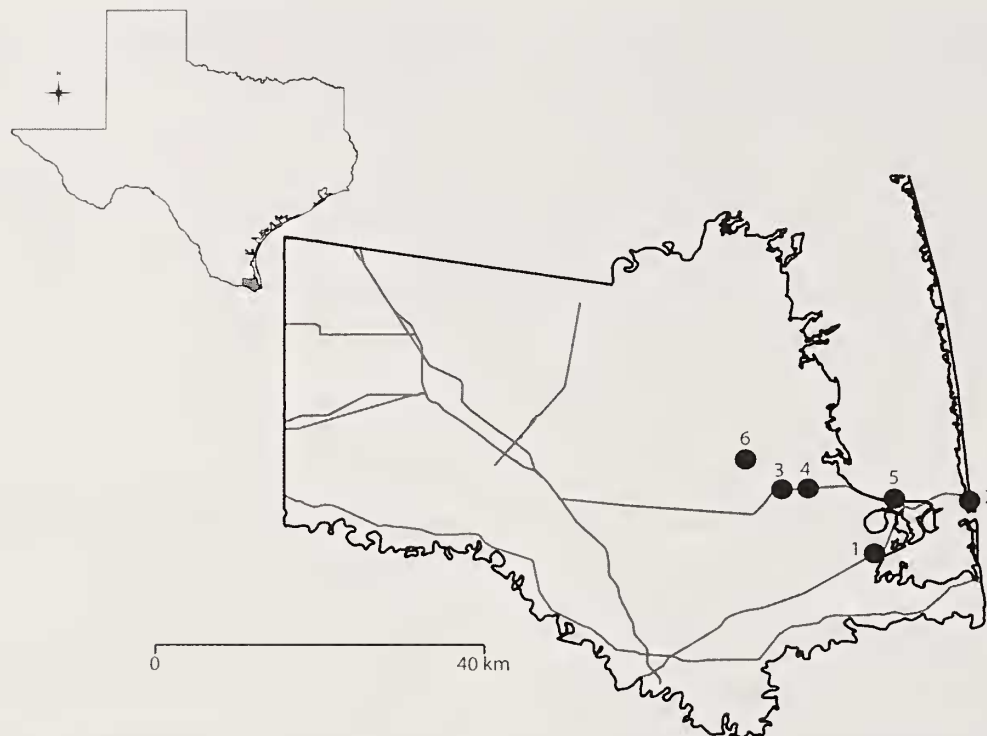


Figure 1. Map with sampling sites for Cameron County populations included in the molecular and soil analyses (Table 1 and Figure 2). The location of Cameron County Texas is depicted in gray in the inset map. State highways are shown as gray lines in the Cameron County map.

species under the phylogenetic species concept (PSC). Under the PSC, species are both the smallest units for which phylogenetic relationships can be reliably inferred (Baum and Shaw, 1995) and entities residing at the transition between evolutionary relationships that are best reflected as reticulate genealogical connections (Goldstein et al., 2000). With the Species Delimitation Plugin (SDP) (Masters et al., 2011) in Geneious 8 (Biomatters Ltd., Kearse et al., 2012), we calculated Rosenberg's $P_{(AB)}$ to test the reciprocal monophyly of each labeled clade and its closest clade (Rosenberg, 2007). Rejection of the null hypothesis suggests genealogical separation of distinct taxa versus monophyly arising randomly according to a Yule model. Significance was determined following Rosenberg (2007). The SDP also assessed the probability of assigning a given individual to its member clade in two ways (Ross et al., 2008). A strict probability was determined for placing an individual into the correct clade while not placing it into the sister clade, and a liberal probability was calculated for placing an individual into either the correct clade or sister clade (Masters et al., 2011). We also calculated the genealogical sorting index (GSI) for each labeled clade in our phylogeny. The GSI statistic quantifies the degree of exclusive ancestry for identified groups in a rooted tree and tests whether it is greater or less than that expected by chance. Significant results suggest that *a priori* groups do not represent a single mixed genealogical ancestry. We employed a GSI web service (<http://molecularevolution.org/software/phylogenetics/gsi/>) and assessed significance at $\alpha=0.05$

using 10,000 tip label permutations on our fixed topology. Finally, we used the Poisson tree processes (PTP) model (Zhang et al., 2013) to assess whether the number of substitutions between our labeled clades was significantly higher than that within those same clades; significantly more substitutions between clades implies that they represent separate phylogenetic species. This method does not require an ultrametric phylogeny nor an evolutionary time context. We used an online likelihood implementation of PTP (<http://species.h-its.org/ptp/>) with default settings.

Morphological Examination: We collected five measurements from each of 42 individuals, measured to the nearest 0.1 mm with digital calipers: maximum shell height parallel to the axis of coiling, maximum shell width perpendicular to the axis of coiling, maximum aperture width, aperture height perpendicular to aperture width measurement, and maximum umbilicus diameter. Only adult specimens with complete reflected lips were measured. The number of whorls was estimated using the method described by Cheatum and Fullington (1971: 15, fig. 1c) to the nearest 0.25 of a whorl at 20 \times magnification. This method counts each whorl as a complete spiral turn of the shell. A stacked composite image of the holotype shell was assembled using Helicon Focus 6.7.1 (Helicon Soft Limited). To relax snails for dissection, snails were drowned in room temperature water for 30 minutes, followed by incubation for 90 minutes at 37 $^{\circ}$ C (Kruckenhauser et al., 2011). Following relaxation snails were preserved in 70% ethanol until dissection. Soft tissues

Table 1. Locality and collection information for populations examined in this study for molecular analysis, soil analysis, and additional material examined. #Included from Perez 2011. *At this site only dry shells of *P. salina* were present. Specimens are deposited at the Academy of Natural Sciences of Philadelphia Drexler University (ANSP). ANSP numbers beginning with “A” are lots preserved in alcohol. Field Museum of Natural History accession is coded FMNH. Latitude and longitude presented in decimal degrees. Cameron County Population Numbers are those labeled on Figure 1.

Species	Cameron County Population Number	Collection information	Museum Numbers	Lat.	Long.
<i>Records for Molecular Analyses</i>					
<i>Praticolella salina</i>	1	8 km S of Port Isabel, Highway 48, Entrance to Laguna Atascosa National Wildlife Refuge, Cameron Co., Texas, K.E. Perez, E. Ruiz, 8 Nov 2014. Type locality.	ANSP A24736 (Holotype), ANSP 467509 (Paratypes), ANSP A24737	25.9957	−97.311
<i>Praticolella salina</i>	2	University of Texas Rio Grande Valley Coastal Studies Lab, Isla Blanca Park, S end of South Padre Island, Cameron Co., Texas, K.E. Perez, D. Deshommes, 19 Oct 2014.	ANSP 467487, A24735	26.0755	−97.159
<i>Praticolella salina</i>	3	2.5 km W of the water treatment facility at Laguna Vista. S side of Highway 100, Cameron Co., Texas, K.E. Perez, D. Deshommes, 19 Oct 2014.	ANSP 467486, A24734	26.0903	−97.348
<i>Praticolella salina</i>	4	1 km W of Laguna Vista, S side of Highway 100, Cameron Co., Texas, K.E. Perez, E. Ruiz, 8 Nov 2014.	ANSP-A 24738	26.0904	−97.349
<i>Praticolella salina</i> (called “ <i>P. griseola</i> Cameron Co.” in Perez 2011)	5 [#]	Port Isabel High School on N (bay side) of Park Road 1, Port Isabel, Cameron Co., Texas, T. Glenn Littleton and N.E. Strenth, 18 Dec 1990.	ANSP-A 22074, ANSP 426021	26.077	−97.227
<i>Additional location for soil analysis</i>					
<i>Praticolella salina</i>	6*	Bayview, Cameron Co. Texas, Toronja Dr., 100 m from Farm-to-Market Road 2480, K.E. Perez, E. Ruiz, 8 Jul 2015.	ANSP 456531	26.119604	−97.400126
<i>Additional Material Examined</i>					
<i>Praticolella salina</i>		2.5 km W of the water treatment facility at Laguna Vista, S side of Highway 100, Cameron Co., Texas, K.E. Perez, E. Ruiz, 8 Jul 2015.	ANSP 456530, A24738	26.0903	−97.348
<i>Praticolella salina</i>		Bayview, Cameron Co., Texas, Toronja Dr., 100 m from Farm-to-Market Road 2480, K.E. Perez, E. Ruiz, 8 Jul 2015.	ANSP 456531	26.119604	−97.400126
<i>Praticolella salina</i>		2.5 km W of Laguna Vista on S. side of Highway 100, Cameron Co., Texas, E. Ruiz, 29 Mar 2016.	ANSP 456532, A24739	26.0903	−97.348
<i>Praticolella salina</i>		8 km S of Port Isabel, Highway 48, Entrance to Laguna Atascosa National Wildlife Refuge, Cameron Co., Texas, K.E. Perez, E. Ruiz, 8 Jul 2015.	ANSP 467533	25.9957	−97.311
<i>Praticolella salina</i>		14.5 km W of Boca Chica, Brady Unit, Laguna Atascosa National Wildlife Refuge, off HWY 4. Cameron Co., Texas, K.E. Perez, E. Ruiz, 30 Oct 2014.	ANSP 467534	25.9621	−97.27893
<i>Praticolella mexicana</i>		Mcallen, TX, 600 N. 7th St., backyard, Hidalgo Co., Texas, K.E. Perez, 4 Aug 2015.	ANSP A24740	26.2085	−98.2255
<i>Praticolella salina</i> (labeled as <i>P. griseola</i>)		Port Brownsville, Cameron Co., Texas, L. Hubricht, 9 Sep 1954.	FMNH 259156	25.9509	−97.4109

were removed from the shell and a mid-sagittal incision was used to expose the internal anatomy. Connective tissue was removed followed by separation of the genitalia. All structures were photographed in water.

Soil Sampling: To determine the soil salinity in the habitat of this species, soil samples were collected at four of the collection localities for live snails from Cameron County (Sites 1, 2, 3, and 5 from Table 1). Small samples of soil were collected by a gloved hand covering the entire local extent of the population into a single, 5 L collection. These samples were mixed in a plastic bucket and a subsample was sent to the University of Louisiana at Monroe Environmental Analysis Laboratory for quantification of all extractable elements. Soil descriptors followed Soil Survey Division Staff (1993).

RESULTS

Twenty new 16S sequences (GenBank KX431997–KX432016) from four populations of the new species *P. salina* were generated. Maximum likelihood analysis of 436 bp of 16S mt sequences of 110 individuals of nine putative species of *Praticolella* yielded a single tree (log likelihood = -4465.5927; Figure 2) with an overall tree topology similar to that found by Perez (2011). Outgroups included in the analysis were representatives of the other genera of Polygyrini included in Perez (2011): *Lobosculum pustuloides* (Bland, 1858); *Polygyra septemvolva* Say, 1818; *Polygyra cereolus* (Mühlfeld, 1816); *Daedalochila hippocrepis* (Pfeiffer, 1848); *Linisa texasiana* (Moricand, 1833); and *Millerelix mooreana* (Wi.G. Binney, 1858). Species-level clades were well supported but relationships among these taxa had little bootstrap support. Two putative species-level clades (monophyletic groups identified by the species delimitation analyses conducted) we recognized currently lack names: an unnamed species from Soto de la Marina, Tamaulipas, Mexico, herein referred to as “Soto”; and an unnamed species from an introduced (greenhouse) population in Florida, USA, herein referred to as “Florida”. Three other nominal species (*P. taeniata* Pilsbry, 1940; *P. pachyloma* (Menke in Pfeiffer, 1847); and *P. candida* Hubricht, 1983) appeared to form a single species-level clade from South Texas, referred to herein as the “South Texas Clade”. A weakly supported clade (54%) suggested a close relationship between *P. salina* and the Florida population. The *P. salina* clade had some internal population-level molecular structuring with individuals from each population appearing in the various shallow clades with the exception of the South Padre Island individuals which are separate.

We tested our nine labeled species-level clades (Figure 2) using three species delineation methods. Based on Rosenberg's $P_{(AB)}$, the SDP supported recognition of seven of our nine labeled clades as reciprocally monophyletic taxonomic entities (Table 2 and Figure 2). The *Praticolella berlandieriana* clade and South Texas Clade had non-significant $P_{(AB)}$ values. Probabilities of

assigning individuals to their correct clades varied from 59–95% under the “strict” method and 87–99% under the “liberal” method. The clade representing the new species had probabilities of 92% and 99% under the “strict” and “liberal” criteria respectively. All nine labeled clades possessed significant GSI values ($p < 0.05$), suggesting no evidence of mixed ancestry in any group. The maximum likelihood PTP solution identified six of our labeled clades as possible phylogenetic species: *P. berlandieriana*; *P. trimatrix*; South Texas Clade; Soto; Florida; and *P. salina*. These species were supported by all three species delineation methods, however, *P. mexicana*, *P. griseola*, and *P. flavescens* were not supported by PTP, perhaps because of unequal sampling or unrecognized diversity in these clades.

Soils at Site 1 (type locality) had a pH of 7.67, a salinity of 11.6 parts per thousand (ppt), and contained 0.89% organic matter. Across all sites, pH ranged from 7.13 to 8.33, salinity from 0.42 to 22.9 ppt, and organic matter from 0.13% to 1.59%. This indicated that *P. salina* was collected in areas with neutral to moderately alkaline mineral soils. The salt marsh sites (sites 1 and 3) were considered highly saline, while the dune (site 2) and agricultural (site 6) sites were considered non-saline.

SYSTEMATICS

Class Gastropoda Cuvier, 1791
Family Polygyridae Pilsbry, 1930

Genus *Praticolella* von Martens, 1892

Dorcasia Binney, 1878: 356.

Praticola Strebel and Pfeiffer, 1880: 38 [non Swainson, 1837]

Praticolella von Martens, 1892: 138.

Type Species: *Praticola ocampi* Strebel and Pfeiffer, 1880 (= *Helix ampla* Pfeiffer, 1866)

Praticolella salina new species Perez and Ruiz, 2017 (Figures 4–11)

Helix griseola Pfeiffer, 1841.—Binney, 1857: pl. 49 fig. 2, pl. 72 fig. 20.

Praticolella griseola (Pfeiffer, 1841).—Pilsbry, 1940: 690 (misidentification in part), fig. 425; Webb, 1951: 140, pl. 48 fig. 30; Rehder, 1966: 290–291 (misidentification in part), fig. 20; Cheatam and Fullington, 1971: 38–39 (misidentification in part), figs. 2, 12. Neck, 1977: 1–4 (misidentification in part).

Diagnosis: Peristome reflected without inner thickening and narrow throughout, unique among Texas *Praticolella*; lower surface of body whorl brown with a single to several white bands; shell wider than high.

Description: Shell large for *Praticolella*, narrowly umbilicate, depressed, brown with white pigmented stripes. Peristome mostly white, barely reflected at parietal wall but heavily reflected at umbilicus, partially obstructing

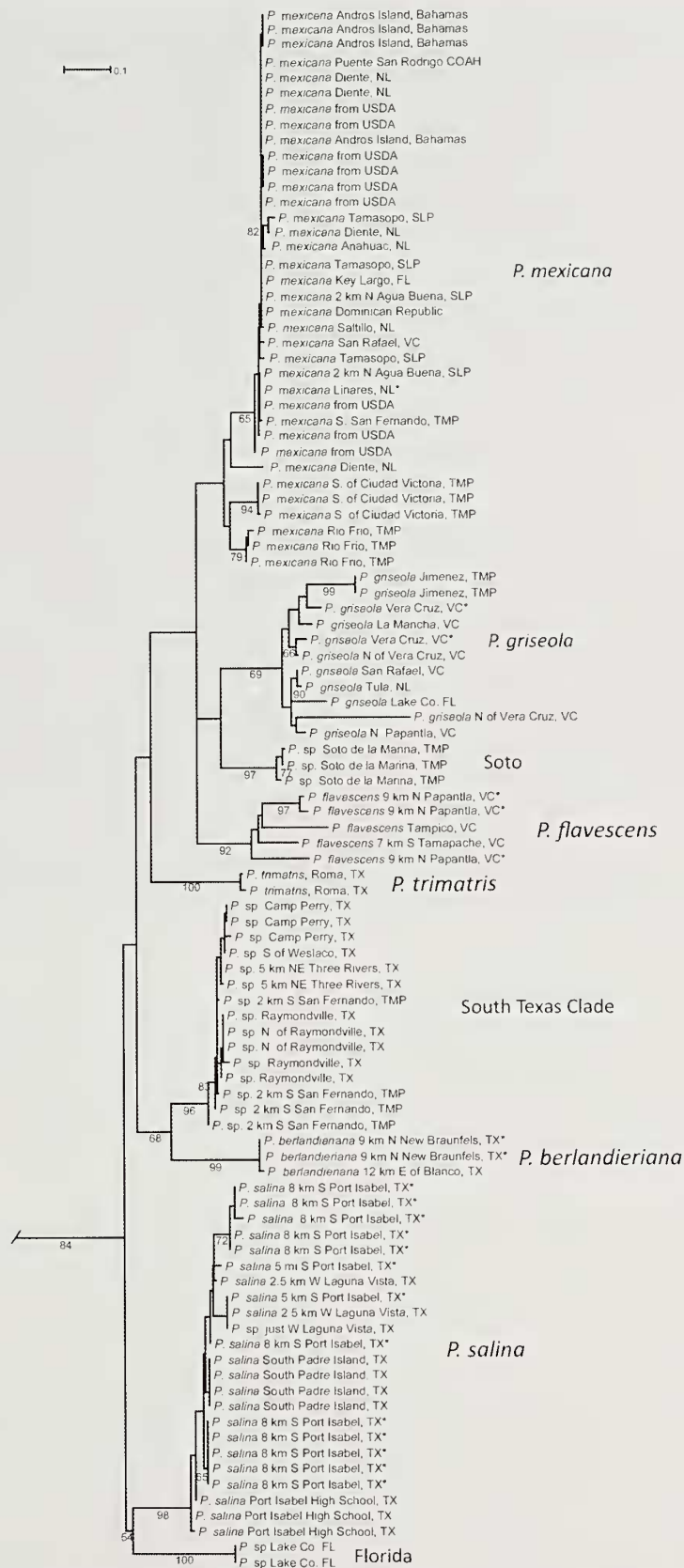


Figure 2. Maximum likelihood phylogeny based on 436 bp of 16S mt sequences of 110 individuals. Only *Praticolella* sensu stricto are shown. Bootstrap values >50% shown below the nodes. Individuals marked with * were collected from type locality. Outgroups are omitted from the figured tree.

Table 2. Results from the Species Delimitation Plugin analysis. Clades correspond to those in Figure 2. Ps and Pl are probabilities of correct identification under strict and liberal criteria respectively. Asterisks (*) signify significant values of Rosenberg's $P_{(AB)}$ and thus separate taxonomic entities by that measure. Clades with significant GSI values and identified as possible phylogenetic species by PTP are also indicated.

Labeled Clade	Ps	Pl	$P_{(AB)}$	GSI	PTP
<i>P. salina</i>	0.92	0.97	$2.7 \times 10^{-1} *$	yes	yes
Florida	0.59	0.98	$2.7 \times 10^{-1} *$	yes	yes
South Texas Clade	0.95	0.99	1.4×10^{-1}	yes	yes
<i>P. berlandieriana</i>	0.78	0.99	1.4×10^{-1}	yes	yes
<i>P. trimatrix</i>	0.58	0.97	$4.5 \times 10^{-5} *$	yes	yes
<i>P. mexicana</i>	0.94	0.98	$1.0 \times 10^{-2s} *$	yes	no
Soto	0.77	0.99	$4.2 \times 10^{-1} *$	yes	yes
<i>P. griseola</i>	0.74	0.92	$4.2 \times 10^{-1} *$	yes	no
<i>P. flavescens</i>	0.61	0.87	$2.6 \times 10^{-10} *$	yes	no

umbilicus in most individuals. Aperture slightly lunate with light parietal callus. Suture smooth but uneven where intersected by growth lines. Protoconch smooth, with longitudinal growth lines (radial lines) appearing by the second spire whorl. Spire and body whorls white above a single translucent, light-brown band around the periphery; up to six additional white stripes below that translucent band. Umbilicus outlined by a single translucent, light-brown band often followed by a white pigmented band. Mean shell height 9.10 ± 0.48 mm, width 12.21 ± 0.68 mm, height/width ratio 0.75; mean aperture height 6.4 ± 0.66 mm, width 6.42 ± 0.14 and height/width ratio 0.88 (Table 3).

Body color brown in life. Largest branch of divided penial retractor muscle inserted on apex of penis. Two smaller branches attached to penis with vas deferens passing between them (Figure 3). Vas deferens of consistent diameter across its length. Penis bipartite with one smooth bulb and distinct appendix. Penial appendix, in the unextended state, slightly narrower at penial attachment, widening and becoming bulbous, about one-half total penial width. Distal end of the penial appendix slightly hooked. Epiphallus noticeably smaller in diameter than the penis, with the vas deferens at the terminal end; flagellum absent. Bursa copulatrix thin, clavate, widening slightly at the terminus. Ovotestis appears as a sponge-like, irregular mass.



Figure 3. Internal anatomy of specimen from Cameron County, TX. ANSP A24739. AG, albumen gland; BC, bursa copulatrix; C, carrefour (spermatheca and fertilization pouch complex); EP, epiphallus; G, genital pore; HD, hermaphroditic duct; OT, ovotestis; P, penis; PA, penial appendix; PRM, penial retractor muscle; SO, spermoviduct; V, vagina; VD, vas deferens.

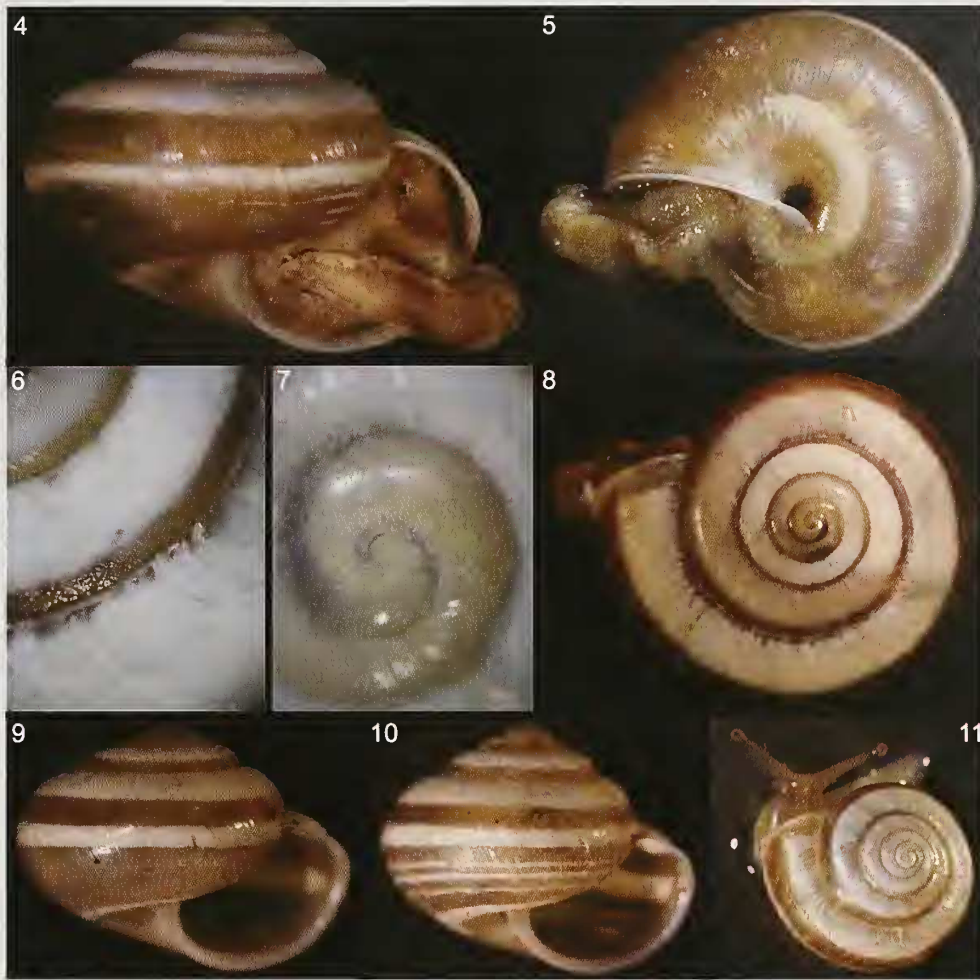
Type Material: Holotype, ANSP A24736; Paratypes, ANSP 467509 (35 individuals), all from type locality.

Type Locality: 8 km south of Port Isabel on HWY 48, Laguna Atascosa National Wildlife Refuge, Cameron County, TX. 25.9957 N, -97.311 W, (8 November 2014, coll. K. E. Perez and E. Ruiz).

Distribution and Habitat: Vegetated dunes and sands and clay soils on South Padre Island and coastal Cameron County, Texas. These locales are associated with Gulf Coast saline prairie habitats in the South Texas Lomas

Table 3. Shell measurements for the *Praticolella* species under consideration. Only adult shells with a full lip were measured. Measurements for *P. griseola* (n=36) are from (Perez 2011) and *P. salina* (n=42) from the present study. Values present are the range of values, mean, and standard deviation. Measurements taken: shell height (h), width (w), aperture height (aph), aperture width (apw), umbilicus width (umb), and number of whorls (# of whorls).

Species	h (mm)	w (mm)	aph (mm)	apw (mm)	umb (mm)	# of whorls
<i>P. griseola</i>	8.32–11.29	5.8–7.92	4.4–6.7	4.16–5.75	0.38–1.03	4.75–5.5
	9.65±0.78	6.91±0.51	5.34±0.47	4.88±0.39	0.71±0.16	5.12±0.16
<i>P. salina</i>	8.04–10.28	10.84–13.55	4.65–6.71	5.17–7.84	0.39–0.97	5–5.75
	9.10±0.48	12.21±0.68	5.63±0.50	6.42±0.66	0.65±0.14	5.39±0.18



Figures 4–11. Shells of *Praticolella salina* new species. **4–8.** Holotype, ANSP A24736, from type locality: 8 km S of Port Isabel on HW 48, Laguna Atascosa National Wildlife Refuge, Cameron County, Texas, 8 Nov 2014, K.E. Perez, E. Ruiz, lateral, basal, and apical views of the shell, close up of suture and embryonic whorls, $w=13.60$ mm, $h=10.03$ mm, 5.5 whorls. **9.** ANSP 467487; UTRGV Coastal Studies Lab, Isla Blanca Park, south end of South Padre Island, Cameron County, Texas, 19 Oct 2014, K.E. Perez, D. Deshommes, $w=10.06$ mm, $h=7.64$ mm, 5.0 whorls. **10.** ANSP 467487; UTRGV Coastal Studies Lab, Isla Blanca Park, south end of South Padre Island, Cameron County, Texas, 19 Oct 2014, K.E. Perez, D. Deshommes, $w=10.85$ mm, $h=8.59$ mm, 5.5 whorls. **11.** ANSP A24739, 2.5 km W of the water treatment facility at Laguna Vista, S side of HWY 100, Cameron County, Texas, 29 March 2016, E. Ruiz, $w=11.87$ mm, $h=9.23$ mm, 5.25 whorls.

ecological system (Natureserve, 2016), a rare plant community recognized by Texas Parks & Wildlife. Individuals have been found in Dune sand, Harlingen clay, Point Isabel clay, Lomalto clay, and Laredo silty clay loam soil types. Dominant vegetation in the clay soils includes shoregrass (*Monanthochloe littoralis*), bushy seaside tansy (*Borrchia frutescens*), and Florida / gutta-percha Mayten (*Maytenus phyllanthoides*); all are salt tolerant species. Snails were found crawling or estivating on cactus (*Opuntia* sp.) at Sites 1–4, Site 5 was recently modified to citrus orchards and cornfields, with no cactus present. This species appears to have a very limited distribution that is likely reduced from its previous extent. We find only dry shells of *P. salina* farther inland and in close proximity to extant *P. mexicana* colonies. The species likely extends into coastal, northern

Tamaulipas, Mexico as well, but that area has not been sampled by the authors.

Etymology: From Latin, *salinus*, salty (derivative of *sal*), in reference to the species' unusual occurrence in highly saline terrestrial habitats.

Comparisons with Other *Praticolella*: The shell of *P. salina* is distinct from that of *P. griseola* in being larger, wider and less globose, and lacking a diagnostic cinnamon-brown pigmented band. The aperture of *P. salina* is also wider than high compared to the nearly round aperture of *P. griseola*. *Praticolella salina* can be distinguished from shells of the South Texas *Praticolella* clade members by its thin versus thickened and deeply reflected peristome. It can be distinguished from *P. mexicana* in always

possessing some white pigmented bands that follow the axis of coiling; none of them, however, run against the axis of coiling or have a pattern of alternating white, pigmented and brown, unpigmented, broken "rays" running perpendicular to the axis of coiling as is often the case in *P. mexicana*.

The penial appendix of *P. salina* is distinctive as it is distally clavate, hooked, and about half the width of the penis. In *P. mexicana*, this structure is equally wide along its length, lacks any hook, and is slightly less than the penile width. The bursa copulatrix of *P. salina* is clavate, only slightly wider at the distal end than at the insertion into the vagina. This structure is distinguishable from that of the South Texas clade which is reniform (Vanatta, 1915), and from both *P. mexicana* and *P. berlandtieriana* (Webb, 1967), which have expanded spatulate distal ends that taper to narrow insertion points.

Remarks: Perez (2011) reviewed the turbulent taxonomic history of *Praticolella* from southern Texas and northern Mexico, especially as it relates to nominal *P. griseola* from Cameron County, Texas. *Praticolella griseola* was originally described from Veracruz, Mexico, by Pfeiffer (1841). Orcutt (1915) first considered *P. griseola* of Texas to be distinct instead of an example of a polymorphic species. Pilsbry (1940) figured *P. salina* from Brownsville, Texas, as *P. griseola* and noted that Brownsville specimens were larger and banded differently than the type specimen. Rehder (1966) compared *P. griseola* from throughout its range and considered the Brownsville population to be a distinct race, characterized by large specimens with sharply defined color bands. In their review of Texas *Praticolella*, Cheatum and Fullington (1971) reviewed *P. griseola*. The descriptions, distribution, and measurements given by Cheatum and Fullington for that latter species represent *P. salina* as well as other South Texas species. Neck (1977) revised nomenclatural and distribution records for *P. griseola* of previous authors, and restricted *P. griseola* in Texas to Cameron County near Brownsville and Laguna Atascosa National Wildlife Refuge. Herein we consider all of these treatments of *P. griseola* in South Texas to be consistent with and indicative of *P. salina*.

By restricting its distribution to southern Texas (possibly south to Tamaulipas), we aim to emphasize the separation of *Praticolella salina* and *P. griseola* evidenced by morphological and molecular data. *Praticolella griseola* sensu stricto is a species from the Gulf Coastal Plain of south-central Mexico that has been moved through human activity with established populations in Alabama, Florida, and Louisiana. Any occurrence of true *P. griseola* in Texas is therefore considered an introduction, not a native and/or remnant population. Additional historical records of *P. griseola* in Texas have been or should be reassigned to other species, including *P. mexicana*. As such, we have limited our synonymy to those works that clearly illustrated a shell we consider to be *P. salina*. Other works listing *P. griseola* ambiguously from Texas may represent *P. salina*, but without additional evidence they were excluded.

We often find *Praticolella salina* occurring with other *Praticolella* species. In Brownsville, for example, we confirmed Pilsbry's (1940) observation that it occurs with *P. taeniata*. Similarly, we have also found *P. salina* within a few meters of *P. mexicana*, where the former was in native habitat and the latter in the grassy verge of a roadway. This is reminiscent of how other *Praticolella* species co-occur, such as *P. griseola* and *P. flavescens* in central Mexico.

The present study with extensive sampling in Cameron County found only eight populations of *Praticolella salina* (seven with living individuals present) in a coastal region with rapid habitat modification due to housing and business developments. This finding is typical of land snails, one of the most diverse, relatively poorly known, and imperiled groups of animals globally (Lydeard et al., 2004).

ACKNOWLEDGMENTS

We thank the University of Texas Rio Grande Valley's (UTRGV) Science Education Grant #52007568 funded by the Howard Hughes Medical Institute, ADVANCE Institutional Transformation Grant (NSF# 1209210), UTRGV Faculty Research Council, Undergraduate Research Initiative, and College of Sciences for financial support. This work was supported in part by the National Science Foundation (under grant HRD-1463991). Any opinions, findings, and conclusions or recommendations are those of the authors and do not necessarily reflect the views of NSF. We thank Frank Judd for assistance with plant identification, Zen Faulkes for use of photographic equipment, Patrick Marquez for anatomical photographs, Tim Pearce for a critical review, and Paul Callomon for specimen deposition. We also thank Ned E. Strenth, T. Glenn Littleton, Victoria Garcia Gamboa, Ruth Lopez, Didier Deshommes, and Norma Allie Perez for assistance with collections.

LITERATURE CITED

- Baum, D.A. and K.L. Shaw. 1995. Genealogical perspectives on the species problem. *Experimental and Molecular Approaches to Plant Biosystematics* 53: 123–124.
- Binney, A. 1857. *The Terrestrial Air-breathing Mollusks of the United States and the Adjacent Territories of North America*. Vol. 3. Little, Brown and Company, Boston.
- Castresana, J. 2000. Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. *Molecular Biology and Evolution* 17: 540–552.
- Cheatum, E.P. and R.W. Fullington. 1971. The Aquatic and Land Mollusca of Texas. *Bulletin of the Dallas Museum of Natural History* 1: 1–74.
- Darriba, D., G.L. Taboada, R. Doallo, and D. Posada. 2012. jModelTest 2: more models, new heuristics and parallel computing. *Nature Methods* 9: 772.
- Dereeper, A., V. Guignon, G. Blanc, S. Audic, S. Buffet, F. Chevenet, J.F. Dufayard, S. Guindon, V. Lefort, M. Lescot, J.M. Claverie, and O. Gascuel. 2008. Phylogeny.fr: robust

- phylogenetic analysis for the non-specialist. *Nucleic Acids Research* 36: W465–469.
- Edgar, R.C. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research* 32: 1792–1797.
- Goldstein, P.Z., R. Desalle, G. Amato, and A.P. Vogler. 2000. Conservation genetics at the species boundary. *Conservation Biology* 14: 120–131.
- Guindon, S. and O. Gascuel. 2003. A simple, fast and accurate method to estimate large phylogenies by maximum-likelihood. *Systematic Biology* 52: 696–704.
- Kearse, M., R. Moir, A. Wilson, S. Stones-Havas, M. Cheung, S. Sturrock, S. Buxton, A. Cooper, S. Markowitz, C. Duran, T. Thierer, B. Ashton, P. Mentjies, and A. Drummond. 2012. Geneious Basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* 28: 1647–1649.
- Kruckenhauser, L., J. Harl, and H. Sattmann. 2011. Optimized drowning procedures for pulmonate land snails allowing subsequent DNA analysis and anatomical dissections. *Annalen des Naturhistorischen Museums in Wien, B* 112: 173–175.
- Lydeard, C., R.H. Cowie, W.F. Ponder, A.E. Bogan, P. Bouchet, S.A. Clark, K.S. Cummings, T.J. Frest, O. Gargominy, D.G. Herbert, R. Hershler, K.E. Perez, B. Roth, M. Seddon, E.E. Strong, and F.G. Thompson. 2004. The global decline of nonmarine mollusks. *BioScience* 54: 321–330.
- Masters, B.C., V. Fan and H.A. Ross. 2011. Species delimitation—a geneious plugin for the exploration of species boundaries. *Molecular Ecology Resources* 11: 154–157.
- Natureserve. 2016. NatureServe Explorer: An online encyclopedia of life [web application]. Version 7.0. Retrieved June 28, 2016, from <http://explorer.natureserve.org>.
- Neck, R. 1977. Geographical range of *Praticolella griseola* (Polygyridae): correction and analysis. *The Nautilus* 91: 1–4.
- Neck, R. 1984. Restricted and declining nonmarine mollusks of Texas. Technical Series, Texas Parks and Wildlife Department. 34: 17.
- Orcutt, C.R. 1915. Molluscan World Volume I West American Scientist 20: 208.
- Perez, K.E. 2011. A new species of *Praticolella* (Gastropoda: Polygyridae) from northeastern Mexico and revision of several species of this genus. *The Nautilus* 125: 113–126.
- Pfeiffer, L. 1841. *Symbolae ad Historiam Heliceorum* Vol. 1. Casselis.
- Pilsbry, H.A. 1940. Land Mollusca of North America (north of Mexico) Vol. 1-Part 2. Academy of Natural Sciences of Philadelphia.
- Posada, D. 2003. Using Modeltest and PAUP* to select a model of nucleotide substitution. *Current Protocols in Bioinformatics* 6, Unit 6.5. doi: 10.1002/0471250953.bi0605s00.
- Rehder, H.A. 1966. The non-marine mollusks of Quintana Roo, Mexico with the description of a new species of *Drymaeus* (Pulmonata: Bulimulidae). *Proceedings of the Biological Society of Washington* 79: 273–296.
- Robinson, D.G. 1999. Alien invasions: the effects of the global economy on non-marine gastropod introductions into the United States. *Malacologia* 41: 413–438.
- Rosenberg, N.A. 2007. Statistical tests for taxonomic distinctiveness from observations of monophyly. *Evolution* 61: 317–323.
- Ross, H.A., S. Murugan, and W.L.S. Li. 2008. Testing the reliability of genetic methods of species identification via simulation. *Systematic Biology* 57: 216–230.
- Soil Survey Division Staff. 1993. Soil survey manual. Soil Conservation Service U.S. Department of Agriculture Handbook 18, 315 pp.
- Vanatta, E.G. 1915. A revision of the genus *Praticolella* von Martens 1892. *Proceedings of the Academy of Natural Sciences of Philadelphia* 67: 197–210.
- Webb, G.R. 1967. Erotology of three species of *Praticolella*, and of *Polygyra pustula*. *The Nautilus* 80: 133–140.
- Webb, W.F. 1951. United States Mollusca: a Descriptive Manual of Many of the Marine, Land and Fresh Water Shells of North America, North of Mexico. Ideal Printing, St. Petersburg, Florida.
- Zhang, J., P. Kapli, P. Pavlidis, and A. Stamatakis. 2013. A general species delimitation method with applications to phylogenetic placements. *Bioinformatics* 29: 2869–2876.
- Zwickl, D.J. 2006. Genetic algorithm approaches for the phylogenetic analysis of large biological sequence datasets under the maximum likelihood criterion. Ph.D. dissertation, The University of Texas at Austin.